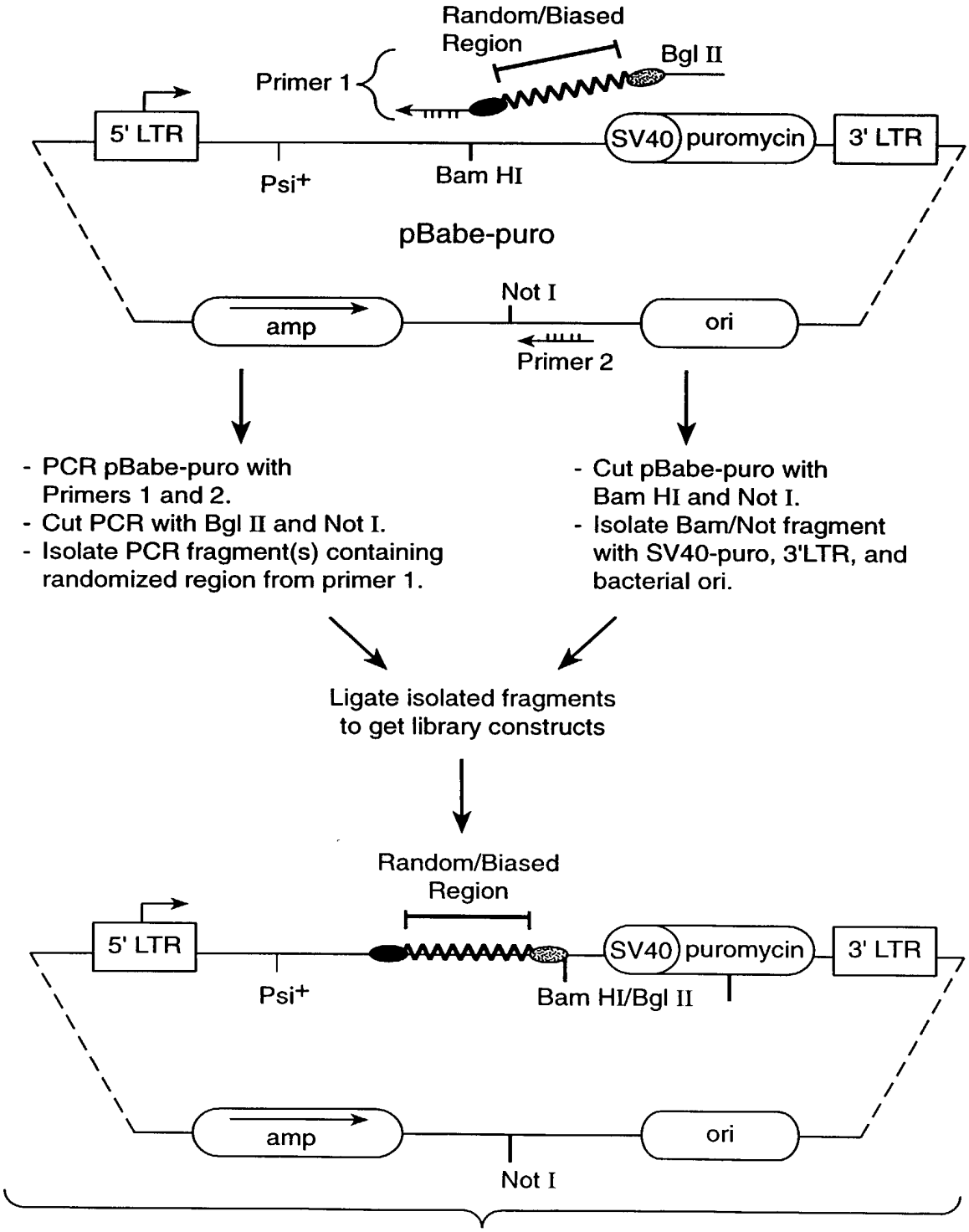

Random/Biased
Region
Bgl II
Primer 1
5' LTR
SV40
puromycin
3' LTR
Psi+
Bam HI
pBabe-puro
Not I
amp
ori
Primer 2
- PCR pBabe-puro with
Primers 1 and 2.
- Cut PCR with Bgl II and Not I.
- Isolate PCR fragment(s) containing
randomized region from primer 1.
- Cut pBabe-puro with
Bam HI and Not I.
- Isolate Bam/Not fragment
with SV40-puro, 3'LTR, and
bacterial ori.
Ligate isolated fragments
to get library constructs
Random/Biased
Region
5' LTR
SV40
puromycin
3' LTR
Psi+
Bam HI/Bgl II
amp
ori
Not I

FIG._1

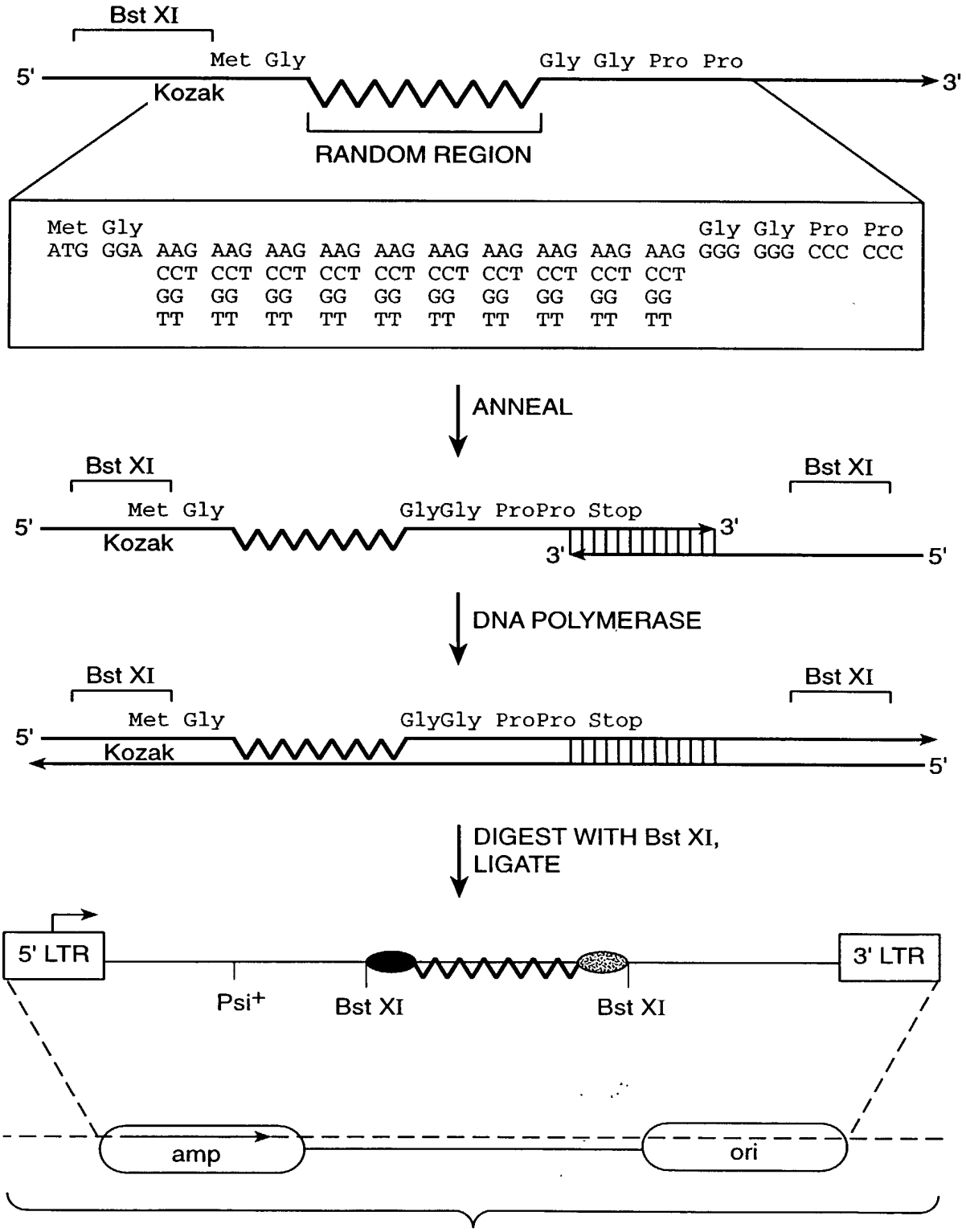

Bst XI
Met Gly
Gly Gly Pro Pro
5'
3'
Kozak
RANDOM REGION
Met Gly
Gly Gly Pro Pro
ATG GGA AAG AAG AAG AAG AAG AAG AAG AAG AAG AAG GGG GGG CCC CCC
CCT CCT CCT CCT CCT CCT CCT CCT CCT CCT
GG GG GG GG GG GG GG GG GG GG
TT TT TT TT TT TT TT TT TT TT
ANNEAL
Bst XI
Bst XI
Met Gly
GlyGly ProPro Stop
5'
Kozak
3'
3'
5'
DNA POLYMERASE
Bst XI
Bst XI
Met Gly
GlyGly ProPro Stop
5'
Kozak
5'
DIGEST WITH Bst XI,
LIGATE
5' LTR
3' LTR
Psi+
Bst XI
Bst XI
amp
ori

FIG._2

FIG._3A

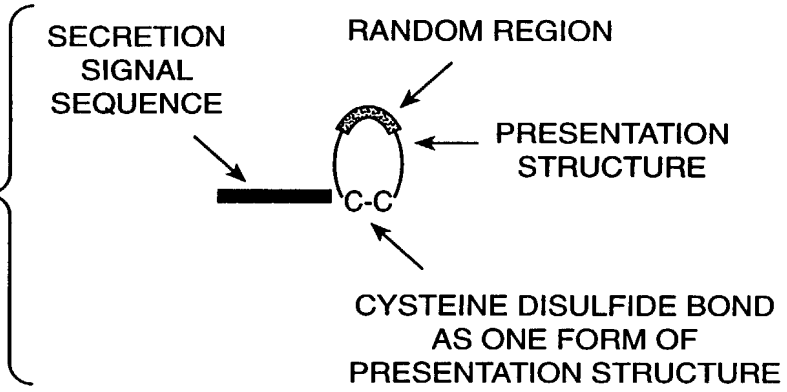


FIG._3B

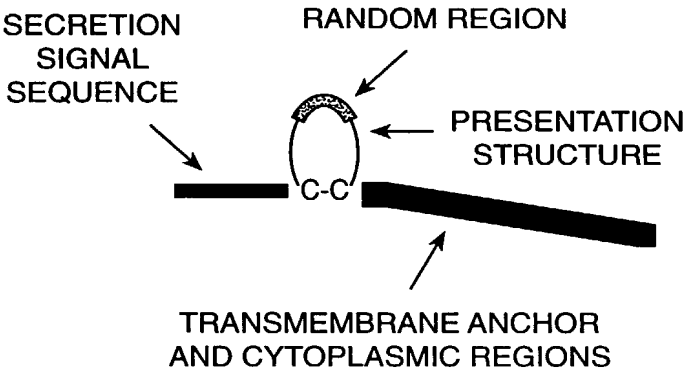
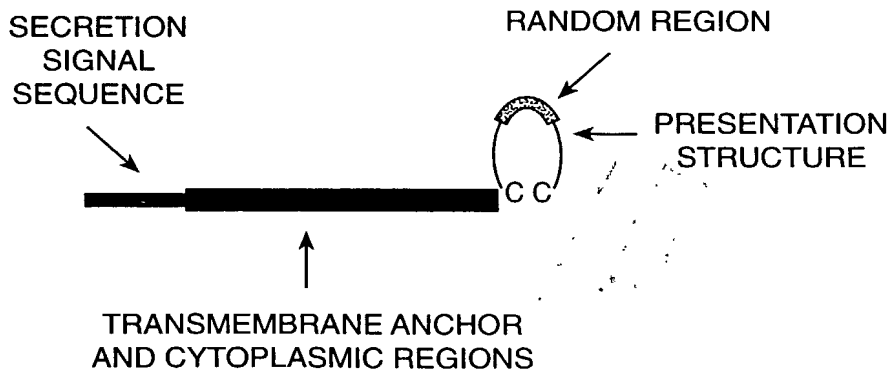


FIG._3C



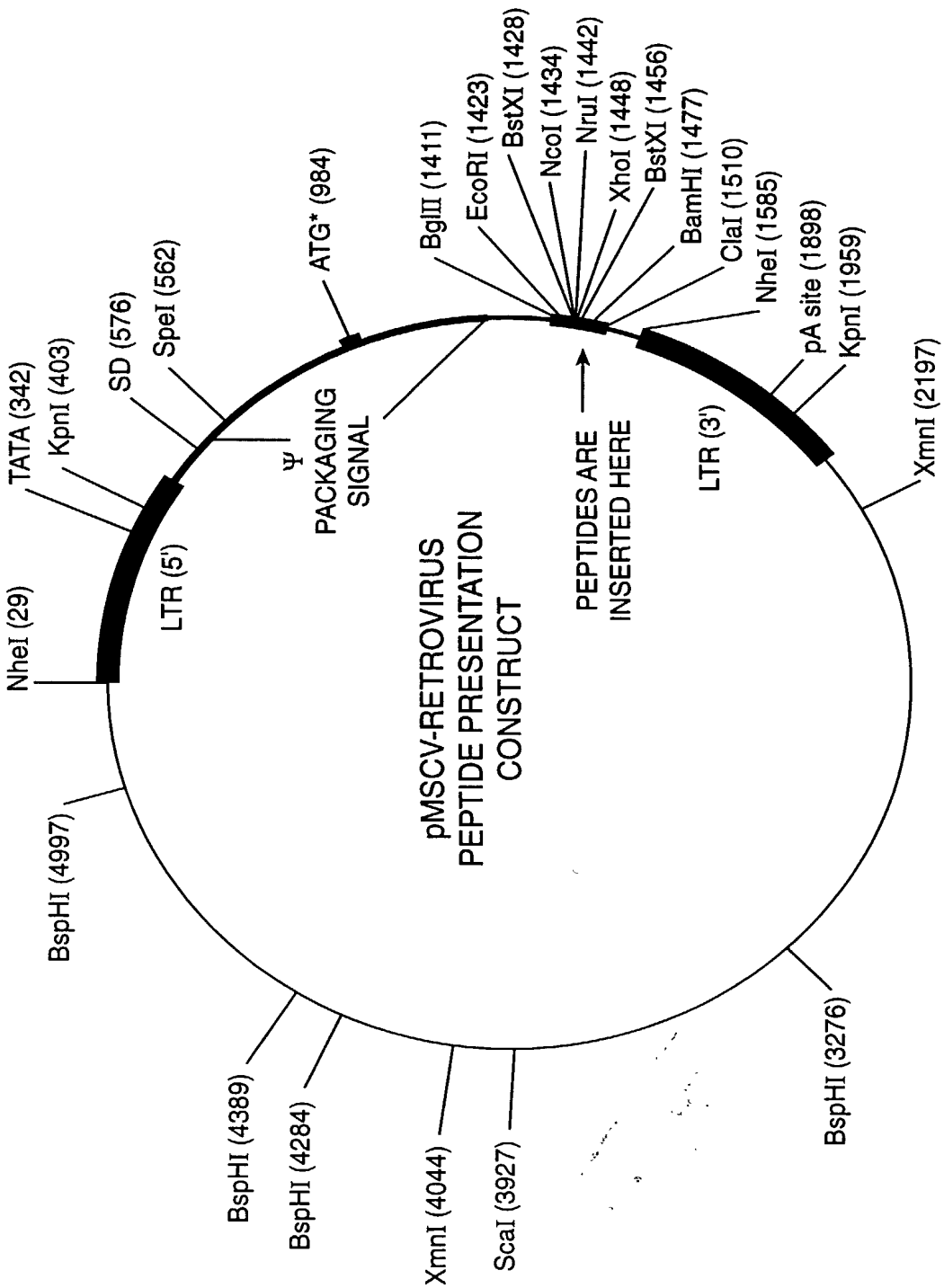

pMSCV-RETROVIRUS
PEPTIDE PRESENTATION
CONSTRUCT
PEPTIDES ARE
INSERTED HERE
LTR (5')
LTR (3')
Ψ
PACKAGING
SIGNAL
NheI (29)
TATA (342)
KpnI (403)
SD (576)
SpeI (562)
ATG* (984)
BglII (1411)
EcoRI (1423)
BstXI (1428)
NcoI (1434)
NruI (1442)
XhoI (1448)
BstXI (1456)
BamHI (1477)
ClaI (1510)
NheI (1585)
pA site (1898)
KpnI (1959)
XmnI (2197)
BspHI (3276)
ScaI (3927)
XmnI (4044)
BspHI (4284)
BspHI (4389)
BspHI (4997)

FIG._4